

45. (Amended) An in vitro method for determining the functional activity of one or more components of the Protein C anticoagulant pathway of the blood coagulation system, comprising:
- (a) providing a blood sample to be analyzed;
  - (b) activating the coagulation cascade by adding a procoagulant reagent to the blood sample to be analyzed;
  - (c) triggering coagulation by adding calcium ions to the blood sample;
  - (d) adding metal ions selected from the group consisting of  $Mg^{+2}$ ,  $Mn^{+2}$ ,  $Zn^{+2}$ ,  $Ni^{+2}$ ,  $Sr^{+2}$ ,  $Cu^{+2}$ , or  $Cu^{+}$ , ions at a concentration that increases the anticoagulant activity of one or more components of the Protein C anticoagulant pathway;
  - (e) incubating a reaction mixture comprising the components recited in steps (a)-(d);
  - (f) observing clotting time; and
  - (g) comparing the clotting time for the blood sample to be analyzed with the clotting time for a normal blood sample as determined by the method recited in steps (a)-(f), thereby allowing for determination of an activity of one or more components of the Protein C anticoagulant pathway.
46. (Amended) The method according to claim 45, wherein the metal ion comprises  $Mg^{2+}$ .
47. (Amended) The method according to claim 46, wherein the metal ion comprises  $Mg^{2+}$  and the amount of the  $Mg^{2+}$  ions added in step (d) is about 20  $\mu$ mol to 10 mmol per liter of reaction mixture.
48. (Amended) The method according to claim 46, wherein the metal ion comprises  $Mg^{2+}$  and the amount of the  $Mg^{2+}$  ions added in step (d) is about 100  $\mu$ mol to 2 mmol per liter of reaction mixture.
49. (Amended) The method according to claim 46, wherein the metal ion comprises  $Mg^{2+}$  and the amount of the  $Mg^{2+}$  ions added in step (d) is about 200  $\mu$ mol to 1 mmol per liter of reaction mixture.
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59. (Amended) The method according to claim 45, wherein the activating the coagulation cascade step occurs via the intrinsic pathway and the procoagulation reagent comprises at least one phospholipid and at least one contact activator.

60. (Amended) The method according to claim 45, wherein the activating the coagulation cascade step occurs via the intrinsic pathway and the procoagulation reagent comprises at least one phospholipid and at least one intrinsic pathway factor selected from the group consisting of Factor IXa, Factor XIIa, and Factor XIa.

61. (Amended) The method according to claim 45, wherein the activating the coagulation cascade step occurs via the intrinsic pathway and the procoagulation reagent comprises at least one phospholipid, at least one contact activator and at least one intrinsic pathway factor selected from the group consisting of Factor IXa, Factor XIIa, and Factor XIa.

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62. (Amended) The method according to claim 61, wherein the at least one contact activator is selected from the group consisting of ellagic acid, collagen, collagen-related substances, and silica.

63. (Amended) The method according to claim 62, wherein the at least one contact activator is a silica selected from the group consisting of micronized silica, colloidal silica, and kaolin.

64. (Amended) The method according to claim 45, wherein the activating the coagulation cascade step occurs via the extrinsic pathway and the procoagulation reagent comprises a material selected from the group consisting of native human tissue factor, recombinant human tissue factor, non-human native tissue factor, non-human recombinant tissue factor, native human Factor VII/VIIa, recombinant human Factor VII/VIIa, native non-human Factor VII/VIIa, and recombinant non-human Factor VII/VIIa.

65. (Amended) The method as in any one of claims 59-61, wherein the at least one phospholipid is selected from the group consisting of synthetic phospholipids, purified phospholipids, and crude extracts of phospholipids derived from biological sources.

66. (Amended) The method according to claim 65, wherein the at least one phospholipid is selected from the group consisting of phosphatidylcholine, phosphatidylserine, and sphingomyelin.

B2 67. (Amended) The method according to claim 45, wherein the activating the coagulation cascade step occurs via the common pathway and the procoagulation reagent comprises a material selected from the group consisting of exogenous Factor Xa, exogenous Factor X and an exogenous activator for Factor X, and an exogenous activator for endogenous Factor X.

68. (Amended) The method according to claim 67, wherein the exogenous activator for Factor X comprises snake venom enzyme.

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70. (Amended) The method according to claim 45, further comprising the step of adding at least one component selected from the group consisting of Protein C, activated Protein C, Protein S, Factor V, Factor Va, a plasma deficient of the Protein C anticoagulant pathway component to be analyzed, and a plasma deficient of all components of the Protein C anticoagulant pathway.

B3 71. (Amended) The method according to claim 45, wherein a fibrin polymerization inhibitor is added to the blood sample to be analyzed.

73. (Amended) The method according to claim 45, wherein the procoagulation reagent comprises a material selected from the group consisting of Factor VIII, Factor VIIIa, Factor IX, Factor X, and prothrombin.

74. (Amended) The method according to claim 45, the method further comprising the step of providing activated Protein C by adding exogenous activated Protein C to the blood sample to be analyzed.

75. (Amended) The method according to claim 45, the method further comprising the step of providing activated Protein C by adding an activator of Protein C to the blood sample to be analyzed.

76. (Amended) The method according to claim 45, the method further comprising the step of providing activated Protein C by adding exogenous Protein C together with an activator of Protein C to the blood sample to be analyzed.

77. (Amended) The method as in any one of claims 74-76, wherein the adding metal ions step occurs simultaneously with the providing activated Protein C step.

78. (Amended) The method as in any one of claims 74-76, wherein the providing activated Protein C step occurs simultaneously with the activating the coagulation cascade step.

79. (Amended) The method as in any one of claims 74-76, wherein the providing activated Protein C step precedes the activating the coagulation cascade step.

80. (Amended) The method as in any one of claims 74-76, wherein the Protein C activator comprises at least one substance selected from the group consisting of Protein C activating snake venom enzyme and thrombin.

81. (Amended) The method as in any of claims 74-76, wherein the Protein C activator comprises thrombomodulin.

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82. (Amended) The method as in any one of claims 74-76, wherein the Protein C activator comprises recombinant Protein C activator.

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83. (Amended) The method according to claim 80, wherein the Protein C activating snake venom enzyme is obtained from the Agkistrodon family.

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85. (New) The method according to claim 83, wherein the snake venom enzyme is obtained from Agkistrodon contortrix contortrix.

86. (New) The method according to claim 83, wherein the snake venom enzyme comprises crude snake venom enzyme.

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87. (New) The method according to claim 83, wherein the snake venom enzyme comprises purified snake venom enzyme.

88. (New) The method according to claim 87, wherein the amount of purified snake venom enzyme is about  $1 \times 10^{-3}$  U to 1 U per milliliter of reaction mixture.

89. (New) The method according to claim 87, wherein the amount of purified snake venom enzyme added is about  $2 \times 10^{-3}$  U to .3 U per milliliter of reaction mixture.

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#### REMARKS

After entry of these amendments, claims 45-57, 59-83 and 85-89 are under consideration. Claims 58 and 84 are hereby canceled without prejudice and without any intention of abandoning the subject matter thereof. Applicants also amend claims 45-49, 59-68, 70, 71, and 73-83 without any intention of disclaiming equivalents thereof. In addition, Applicants have added new claims 85-89. Support for the new and amended claims is found in the specification at least on